temperature of 4°C. Sediment samples collected for sediment bioassay analyses were stored in a refrigerator at 4°C for one to two days after sampling and transported or shipped to the laboratory once sampling was completed.

Laboratory Analysis

All laboratory analyses were conducted by Pacific Eco-Risk Environmental Consulting & Testing (Martinez, Calif.). The methods used in conducting the sediment bioassay evaluations followed established guidelines in *Methods for Assessing the Toxicity of Sediment-Associated Contaminants with Estuarine and Marine Amphipods* (U.S. EPA 1994). Analytical reports are provided in the Technical Appendices, Section II.

Data Analysis

The laboratory performed statistical analyses on the performance of individual sampling areas within monitoring units versus the laboratory control. The control consisted of placing *Eohaustorius* in "pristine" mud collected from a selected reference location -- in this case, Yakima Bay, Oregon. In addition, means and standard errors for each monitoring unit were generated for all three sampling periods. Results were compared with data from the RMP on sediment sampling within San Francisco Bay, specifically San Pablo Bay. As noted in the Sediment Contaminants section, there are two RMP sampling locations in San Pablo Bay: one on the Napa River near Mare Island (BD22) and the other at the southern end of the Petaluma River (BD15). Results were also compared to data available from the Bay Protection and Toxic Cleanup Program. As with the ASC criteria referenced in the Sediment Contaminants section above, the Bay Protection and Toxic Cleanup Program has focused on establishing criteria based on "optimal ambient" or least-polluted conditions. Data from a total of 61 sampling locations were used to develop reference toxicity values or reference envelopes that could be used for comparative purposes (Hunt et al. 1999). A "p" value of 0.10 was selected (as recommended in Hunt et al. 1999), meaning that samples with percent survival below that recorded for a "p" value of 0.10 (in this case, 69.5 percent for *Eohaustorius*) were as toxic or more toxic than the worst 10 percent of the 61 reference sites sampled by the Bay Protection and Toxic Cleanup Program (Hunt et al. 1999).

Food Chain Support and Wildlife Use

Vegetation

Vegetation monitoring was intentionally designed to overlap with the 27 transects established for soil nutrient sampling (Figure 6). Vegetation variables assessed included total vegetation cover; percent vascular plant cover; canopy complexity; percent cover of salt marsh, brackish marsh, and glycophytic plant species; peak standing live and dead crop; and percent cover of vegetation communities (e.g., subsaline seasonal wetland, brackish marsh, etc.). Vegetation monitoring was conducted in 1999 and 2000 (Table 3). However, by the time sampling was conducted in 1999 (late June/early July), vascular plants and micro- and macro-algae within some of the vegetation communities, specifically moist grassland and seasonal wetland, had already begun to senesce or had senesced, complicating monitoring efforts. In 2000, monitoring was conducted between April through June and represented each of the vegetation communities at peak biomass. Therefore, with a few exceptions, only vegetation data from 2000 was used in analyses.

Within the Enhancement Wetlands, monitoring was conducted at 15, 60.6-m (200-foot) long transects established by the original consultants, Jones & Stokes Associates, at representative locations throughout the management units. Monitoring was performed at these transects between 1990 and 1996 by both Jones & Stokes Associates (1990-1991) and Kirven Associates (1993-1996) as part of the mitigation monitoring. Twelve (12) additional 60.6-m (200-foot) long transects were established in the Upland Ponds and the CDFG units in 1999 for a total of 27 transects (Figure 6). Sampling was conducted in the following monitoring units: Reclaimed Water, Reclaimed Water + Muted Tidal, Muted Tidal, Passive Hydrologic Management, Groundwater Pond, Diked Marsh, Seasonal Pond, and Undiked Marsh (Table 4; Figure 6).

Sampling Methodology

Vegetation cover was assessed using a variation of the point-intercept transect method described in *The Manual of California Vegetation* (Sawyer and Keeler-Wolf 1996). At each sampling location, a belt transect approximately 1.8 m (6 feet) in width and 60.6-m (200-feet) long was established. Cover was assessed at 0.6-m (2-foot) increments along the 60.6-m (200-foot) line transect, which bisected the center of the 1.8-m (6-foot) wide belt transect. At each 0.6-m (2-foot) increment, a point was projected vertically onto the ground, and a "hit" was recorded for each element intercepted by the projected point, including vascular plant species, macro-algae, micro-algae, detritus, and standing water. If no cover of any of these elements was present, the point was recorded as bare ground. In addition, any species that was not recorded during vegetation sampling, but was present in the belt transect was also noted. In transects where either macro- or micro-algae were present, representative samples were collected for later identification. Collection of benthic micro-algae samples involved inserting a modified 50-cc syringe into the upper sediment layers where benthic micro-algae appeared to be present. The syringes were wrapped immediately with foil and kept on ice and out of sunlight until delivered to the consultant for analysis.

Vegetation monitoring also incorporated an assessment of standing peak crop. Standing peak crop was sampled at three stratified random locations along the 60.6-m (200-foot) belt transect using a 10-dm^2 quadrat. At each randomly selected location, a left or right direction was randomly chosen, and the quadrat was placed approximately 0.91 m (3 feet) from the line transect. All biomass within the quadrat was harvested, including the season's vascular plant "crop" and micro- and macro-algae. In addition, any detritus or humus present that was not incorporated into the surface soil layers was also collected for separate assessment. Within a soil profile, these detrital layers are often referred to as the 01 and 02 Organic Horizons and are composed of layers of visible vegetative matter and layers of unrecognizable component matter, respectively. Collection was considered complete when only bareground was present within the quadrat. The collected material was then air dried to a constant weight. After the material dried, it was weighed, and the weight was recorded in grams.

Laboratory Analysis

Identification of macro- and micro-algae was performed by Natalie Cosentino-Manning (Cosentino Consulting, Santa Rosa, Calif.).

Data Analysis

Total percent cover or extent of vegetation along the transect was calculated. In addition, a second parameter, vascular plant cover, factored in the potential for presence of several vascular plant species within the "layers" or shrub or herb/forb strata at each intercept point. Due to this "layering" effect, percent cover for this variable could exceed 100 percent. A third parameter, canopy complexity, advanced this concept one step further by incorporating detritus and other primary producers such as macro- and micro-algae and therefore could also exceed 100 percent cover. Calculation of the percent of transect covered by salt marsh, brackish marsh, and glycophytic plant species was performed using vascular plant cover.

To improve compatibility with other mapping efforts in San Francisco Bay, specifically the San Francisco Bay Estuary Project, a similar classification system was used to characterize vegetation communities within the monitoring units (San Francisco Bay Area Wetlands Ecosystem Goals Project 1997). In general, the Study Area incorporated tidal brackish marsh, non-tidal diked brackish marsh, non-tidal diked subsaline seasonal wetlands, freshwater marsh, and moist grassland. Additional categories were added to reflect vegetation communities either not described or broken out of other categories, specifically diked saline seasonal wetland, panne (unvegetated or very sparsely vegetated areas), and seasonal marsh (seasonally saturated/inundated areas dominated by freshwater emergents, primarily *Eleocharis macrostachya*). For the purposes of this study, saline seasonal wetlands were differentiated from subsaline seasonal wetlands on the basis of soil salinity, with saline seasonal wetland areas having mean soil salinities exceeding 1-2 ppt. Also, unlike subsaline seasonal wetlands, which are typically non-tidal, saline seasonal wetlands occurred in both non-tidal and muted tidal areas.

Means and standard errors were calculated for each monitoring unit. Principal Components Analysis (SYSTAT 8.0, SPSS Inc., Chicago, Ill.) was used to informally explore the distribution of sampling locations in relation to vegetation variables. Three principal components were retained for analysis. Rotation did not enhance interpretation of the components.

Zooplankton

Zooplankton sampling focused on characterizing the aquatic invertebrate community within the water column. Variables assessed included total density, species richness, species diversity (Shannon-Wiener's Diversity Index, H'), and community composition. As with some other variables, sampling periods were generally intended to reflect densities and community composition during periods of summer low water; managed flooding during the fall; and winter rain-associated flooding in the spring (Table 3). Sampling occurred in the following monitoring units: Reclaimed Water, Reclaimed Water + Muted Tidal, Muted Tidal, Passive Hydrologic Management, Groundwater Pond, and Undiked Marsh (Table 4). A maximum of 22 locations were sampled depending on water conditions, with fewer samples collected during the summer when some shallow water areas were dry (Figure 8). The invertebrate samples were collected from a variety of habitats, including sloughs/creeks, channels/borrow ditches, ponds, and shallowly ponded areas (e.g., flooded pannes).

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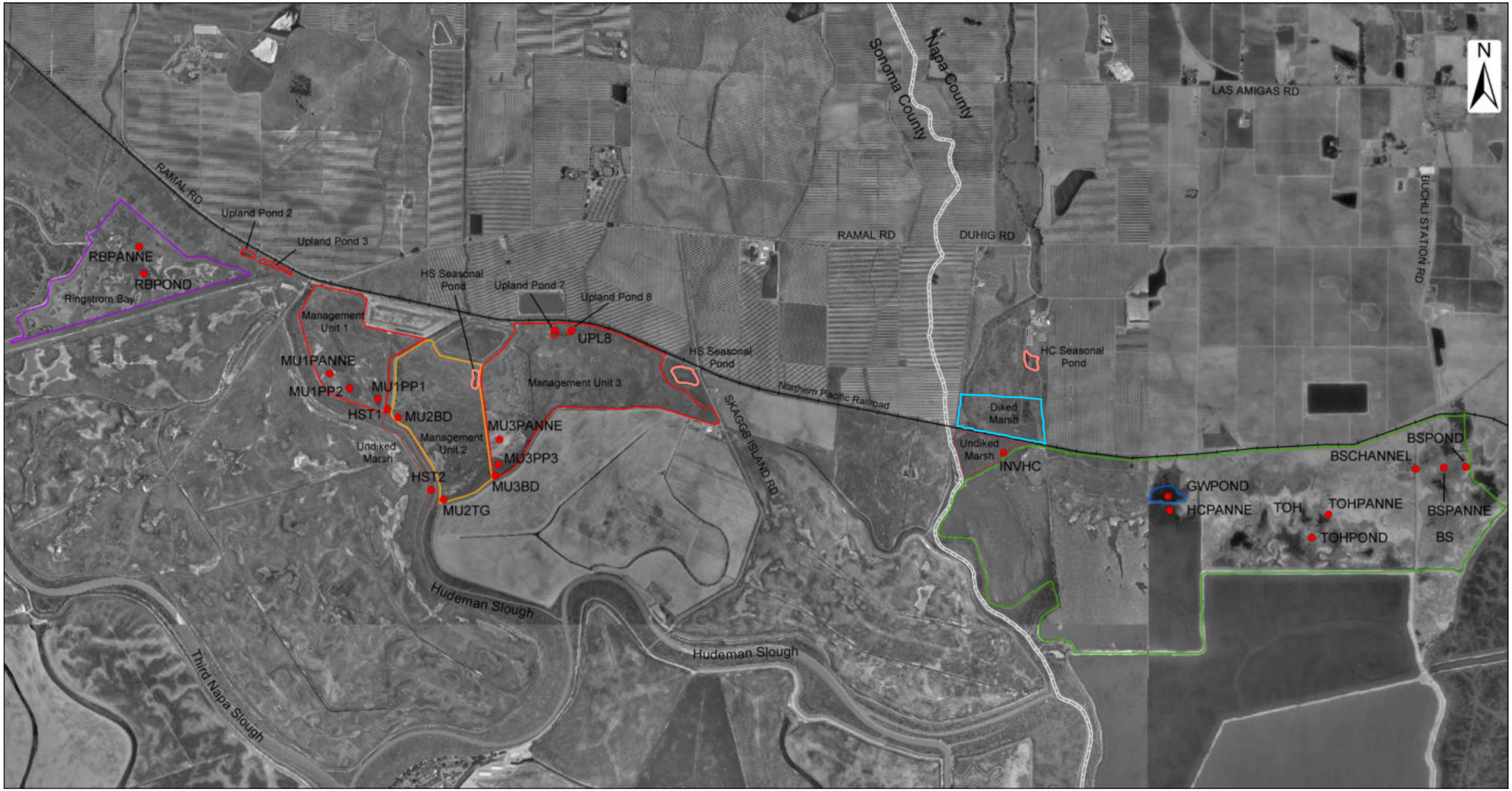

SONOMA COUNTY WATER AGENCY

● Zooplankton and benthic invertebrate sampling locations

Dates of Photos:
July 1993 (Napa County)
June 2001 (Sonoma County)

Monitoring Unit Type

Hydrologically Managed
- Reclaimed
- Reclaimed & Muted Tidal
- Muted Tidal
- Passive
- Groundwater

Hydrologically Unmanaged
- Undiked Marsh
- Diked Marsh
- Seasonal Pond

Figure 8.
Zooplankton and benthic invertebrate sampling locations within the Hudeman Slough Enhancement Wetlands Case Study area.

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To determine what effect the acidic water conditions documented in certain areas might have on zooplankton densities, abundance was compared between sampling locations with varying levels of water pH and alkalinity.

Sampling Methodology

Zooplankton communities were assessed by conducting vertical tows using a 63-µm mesh plankton Nitex net (Turtox; Wildlife Supply Company; Saginaw, Michigan) with a weighted bottom. In channels and ponds, tows were typically conducted by attaching the net to a 5-foot pole and sampling either off the side of channel banks or in the center of ponds using a float tube to minimize sediment disturbance. Use of the float tube and pole was problematic in shallow areas. In these areas, the net was extended out as far as possible from the sampler's body into waters undisturbed by the sampler. In order to sample the same volume of water, the number of tows conducted varied depending on depth of the water. In deeper waters (>20 cm), four vertical tows were conducted by lowering the bottom-weighted net using the pole until the net opening was flush with the sediment bottom and then raising the net vertically. In shallow areas, anywhere from five (5) to 14 tows were conducted by gently extending the net out from the sampler's body and laying the net opening flush with the sediment surface. When possible, enough sampling effort was conducted to collect at least 300 organisms, although densities in certain areas were so low that 300 organisms could not be collected even with numerous replicate tows. Depth of water column sampled was measured for each replicate tow. After each sampling tow, some of the water from the collector bottle was squeezed out, using the net as a filter. Filtered water from the sampling location was then used to rinse all sides of the net to wash any remaining organisms into the collector bottle. Once rinsing was complete, contents of the collector bottle were poured into a 250-ml jar. Each jar was then fixed with 10 percent formalin in the field, and samples were delivered to a consultant for analysis.

Laboratory Analysis

Processing and identification of the zooplankton samples was conducted by Sean Avent, Romberg Tiburon Center for Environmental Studies (Tiburon, Calif.).

Data Analysis

Vertical tows are primarily used for qualitative analysis of organisms present throughout the water column of a sampling location, but quantitative or density data may be obtained by calculating the volume of water as follows: (Length of tow)(π)(Radius of net opening)^2. In addition to total zooplankton densities, species richness (R) and Shannon-Wiener's Diversity Index (H') were calculated. For each of these variables, means and standard errors were generated for each monitoring unit. In addition, mean density and species richness within monitoring units were evaluated for each sampling period.

Species abundance associations within the zooplankton community were explored using detrended correspondence analysis (DCA) on all 22 sampling locations. Similar to PCA, DCA is a form of multivariate indirect gradient analysis that can examine relationships between species and sites, but DCA is considered to be better capable of handling data with nonlinear and unimodal distributions (McGarigal et al. 2000). To improve analysis, extremely rare species were eliminated prior to running the ordination. Direct gradient analysis was also performed using Canonical Correspondence Analysis (CCA). Canonical Correspondence Analysis

integrates some combination of directly measured environmental variables into the analysis of the relationship between species and sites. Environmental variables included water pH, D.O. (mg/L), temperature (°C), and salinity (ppt). DCA and CCA were performed using PC-ORD 4.17 (McCune and Mefford 1999; MjM Software, Gleneden Beach, Oregon).

Zoo Benthos, Epibenthos, and Benthic Infauna

Benthic coring was conducted to assess the abundance and composition of benthic infauna taxa within monitoring units. Similar to the Regional Monitoring Program, sampling was conducted in the winter and summer (Table 3). Sampling occurred in the following monitoring units: Reclaimed Water, Reclaimed Water + Muted Tidal, Muted Tidal, Passive Hydrologic Management, Groundwater Pond, and Undiked Marsh (Table 4). A maximum of 20 locations were sampled (Figure 8). The invertebrate samples were collected from a variety of habitats, including sloughs/creeks, channels/borrow ditches, ponds, and shallowly ponded areas (e.g., flooded pannes).

Sampling Methodology

At each sampling location, three replicate cores were taken to a depth of 15 cm within a radius of 1 to 2 meters of the sampling point. The three cores were combined to represent one benthic infauna sample, and the total volume and surface area of the composite sample was roughly equivalent to that provided by the 0.05 m^2 Ponar Grab used for sampling benthic invertebrates in the RMP. This sampling methodology is limited by the fact that the Ponar Grab, which is used in open sloughs or bays, removes a larger sample than the corer used in this study and therefore has a greater potential to collect large organisms or organisms with a tendency to cluster. At some sampling locations, the corers encountered an impenetrable hard panne 5 cm below the surface. In these areas, only the top 5 cm was collected for analysis. These hard pannes were found principally at two sampling locations: pannes in Management Units 1 and 3 that are seasonally inundated/saturated. Each replicate core was bagged individually, fixed with 95 percent ethanol, and kept on ice in a cooler until delivery to the processor at the end of the sampling day.

Laboratory Analysis

Processing of benthic infauna samples was conducted by Natalie Cosentino-Manning (Cosentino Consulting, Santa Rosa, Calif.). At the laboratory, samples were sieved through a 0.4 mm screen held over a plastic tub. The water in the tub was then poured back through the screen. Material retained on the screen was placed in labeled sample jars with an accompanying data tag and was preserved with 10 percent buffered formalin. After fixation in formalin for a minimum of five days, the samples were transferred to 70 percent ethyl alcohol. Samples were rough sorted into major taxa under 8X magnification. After rough sorting, all organisms were identified to the lowest taxonomic level possible, enumerated, placed in separate vials, and labeled.

Data Analysis

Species composition was compared between monitoring units. Only qualitative estimates of abundance were produced due to differences in size of benthic cores collected. Abundances were separated loosely into the following categories: low, low to moderate, moderate, moderate to high, and high. Results were also used to compare presence and relative abundance of particular species with benthic indicator criteria recently established by the RMP (Lowe and

Thompson 1999). Lowe and Thompson (1999) developed a list of indicator species that are either contamination intolerant or contamination tolerant and some general guidelines to determine whether benthic assemblages reflect impacted or unimpacted conditions. As the data used for developing these guidelines were taken from open water areas within San Francisco Bay, it is not directly comparable with data from areas that are diked and either subjected to attenuated, muted, infrequent, or no tidal flushing. However, limited comparisons with the Lowe and Thompson (1999) study were made to the extent possible.

Avian Surveys

Biweekly avian counts were conducted on 9, 0.15-hectare (ha) monitoring units from September 1999 through August 2001 (Table 3, Figure 9). Study plots were selected based upon hydrologic regime and habitat type at the Enhancement Wetlands and at monitoring units located in the CDFG complex (Ringstrom Bay, Huichica Creek, and Buchli Station units, Table 4). Hydrologic regimes included Reclaimed Water, Reclaimed Water + Muted Tidal, Muted Tidal, and Seasonal Ponds.

Habitats within the study plots were primarily Open Water and Flooded Wetlands. A total of four Open Water study plots were monitored: two in the Reclaimed Water monitoring unit (OF2 and MU3-9) and two managed as Seasonal Ponds (MU3-10 and DFG-13). The Reclaimed Water study plots consisted of constructed upland ponds that are flooded with reclaimed water. The Seasonal Pond study plots incorporated constructed ponds that flood during the winter naturally from precipitation and upland run-off (MU3-10, DFG-13), and/or creek overflow (DFG-13).

Five Flooded Wetland study plots were monitored: three in the Reclaimed Water monitoring unit (MU1-4, MU1-5, and MU3-8), one in the Reclaimed Water + Muted Tidal monitoring unit (OF1), and one in the Muted Tidal monitoring unit (DFG-15).

Sampling Methodology

The study was designed to focus on waterbirds; however, all species observed within the study plot were counted. Fly-overs were counted separately; only aerial foragers (e.g. swallows) were included in the data analysis. Each 10-minute direct count was performed from a single and consistent location on the study plot. The corners of each study plot were marked with a 1.5-m-high PVC pole. Counts were not performed during heavy fog, winds exceeding 32 kilometers per hour (km/h), or heavy rain. The counts were timed to correspond with high tides as it was assumed that the maximum number of birds using the wetland units would occur during high tides when waterbirds would seek foraging and roosting locations that were not inundated by incoming tides in areas closer to San Pablo Bay.

Data Analysis

Species richness (total number of species detected), species diversity (Simpson's and Shannon-Wiener diversity indices), and avian densities (number of birds/study plot) were compared between study plots, for both the entire study and for periods associated with specific hydrologic regimes. Both Simpson's (1/D) and Shannon-Wiener (H') diversity indices were calculated. Simpson's index is a dominance measure that is influenced by the abundance of the most common species (Magurran 1988). The Shannon-Wiener diversity index provides a

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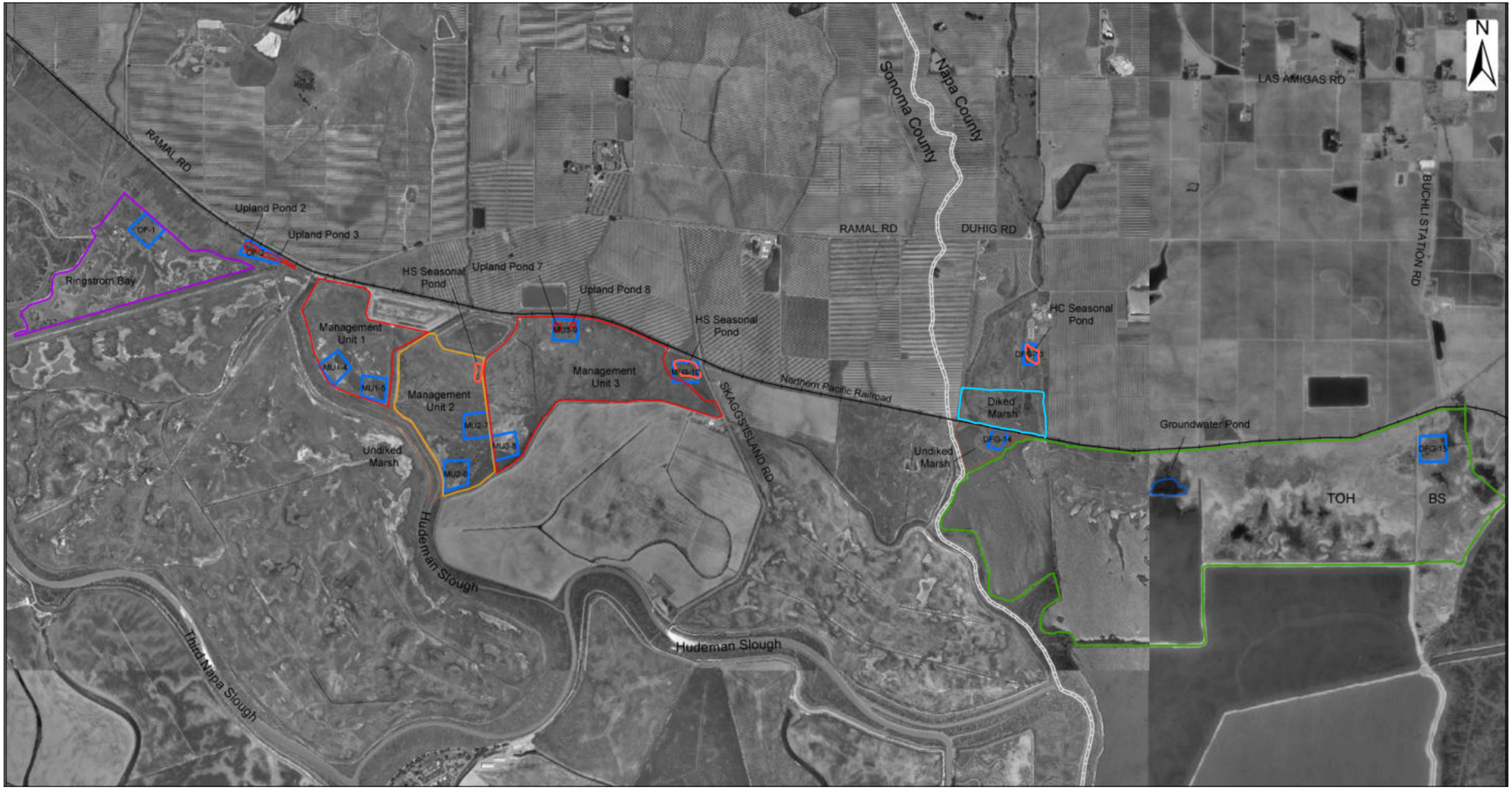

Avian monitoring plots

Monitoring Unit Type

Hydrologically Managed

- Reclaimed
- Reclaimed & Muted Tidal
- Muted Tidal
- Passive
- Groundwater

Hydrologically Unmanaged

- Undiked Marsh
- Diked Marsh
- Seasonal Pond

Dates of Photos:
July 1993 (Napa County)
June 2001 (Sonoma County)

Figure 9.
Avian monitoring plots within the Hudeman Slough Enhancement Wetlands Case Study area.

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measure of species richness and is more affected by less common species (Magurran 1988). Study periods analyzed were September-October, November-April, and May-August. The September-October study period represents the time frame in which the Reclaimed Water, Reclaimed Water + Muted Tidal, and Muted Tidal monitoring units are flooded with either reclaimed water or tidal flows. Between November-April, the Reclaimed Water monitoring units have been drained and, as with the Reclaimed Water + Muted Tidal and Muted Tidal monitoring units, are flooded primarily by precipitation, upland run-off, and creek overflow. The May-August study period represents the dry season, although reclaimed water is maintained year-round in the permanent ponds within the Management Units and two of the Upland Ponds.

Waterbird species were analyzed by foraging guild. Analysis of foraging guilds provides important information regarding food resources and water regimes within waterbird habitat (Weller 1999).

Species richness, species diversity (1/D and H'), and density results for the entire monitoring study (September 1999 through August 2001) were analyzed to determine if there were significant differences between hydrologically managed and unmanaged study plots. Monthly totals were treated as repeated measures. The Kruskal-Wallis non-parametric test was selected as it does not assume normal distribution and allows small sample sizes ($n<50$) (Hintze 2001).

Hudeman Slough study plots MU1-4 and MU1-5 were located within Reclaimed Water monitoring unit MU1. Data collected in these study plots were combined into a single mean (MU1) for each study period because the data could not be considered independent, as the habitats are very similar and undergo identical hydrologic management (flooded simultaneously).

Cluster Analysis

Cluster analysis was used to determine whether the hydrologic management regime classification used in this monitoring study was really the best representation of the structure among sampling locations, if any structure did exist. Sampling locations were pooled into 11 monitoring sub-units, and these sub-units were classified according to 36 biotic and abiotic variables that incorporated water quality, sediment nutrients, vegetation, and zooplankton parameters. Benthic infauna and avian use were not included in the cluster analysis. In addition, a second cluster analysis was run that incorporated six (6) sediment contaminant variables with the 36 biotic and abiotic variables. Euclidean distance and the average linking method of hierarchical clustering produced best clustering results, defined as the least amount of sequentially added small clusters or individual sites that resulted in one or two large clusters. Cluster analysis was performed using SYSTAT 8.0 (SPSS Inc., Chicago, Ill.).

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